Neutrokine-α

```
1    AAATTCAGGATAACTCTCCTGAGGGGTGAGCCAAGCCCTGCCATGTAGTGCACGCAGGAC   60

61   ATCAACAAACACAGATAACAGGAAATGATCCATTCCCTGTGGTCACTTATTCTAAAGGCC   120

121  CCAACCTTCAAAGTTCAAGTAGTGATATGGATGACTCCACAGAAAGGGAGCAGTCACGCC   180
1                                M  D  D  S  T  E  R  E  Q  S  R  L   12

181  TTACTTCTTGCCTTAAGAAAAGAGAAGAAATGAAACTGAAGGAGTGTGTTTCCATCCTCC   240
13    T  S  C  L  K  K  R  E  E  M  K  L  K  E  C  V  S  I  L  P    32
                                                          CD-I

241  CACGGAAGGAAAGCCCCTCTGTCCGATCCTCCAAAGACGGAAAGCTGCTGGCTGCAACCT   300
33    R  K  E  S  P  S  V  R  S  S  K  D  G  K  L  L  A  A  T  L    52
                  CD-I

301  TGCTGCTGGCACTGCTGTCTTGCTGCCTCACGGTGGTGTCTTTCTACCAGGTGGCCGCCC   360
53    L  L  A  L  L  S  C  C  L  T  V  V  S  F  Y  Q  V  A  A  L    72

361  TGCAAGGGGACCTGGCCAGCCTCCGGGCAGAGCTGCAGGGCCACCACGCGGAGAAGCTGC   420
73    Q  G  D  L  A  S  L  R  A  E  L  Q  G  H  H  A  E  K  L  P    92
                  CD-II

421  CAGCAGGAGCAGGAGCCCCCAAGGCCGGCCTGGAGGAAGCTCCAGCTGTCACCGCGGGAC   480
93    A  G  A  G  A  P  K  A  G  L  E  E  A  P  A  V  T  A  G  L   112
                  CD-III
                                    #
481  TGAAAATCTTTGAACCACCAGCTCCAGGAGAAGGCAACTCCAGTCAGAACAGCAGAAATA   540
113   K  I  F  E  P  P  A  P  G  E  G  N  S  S  Q  N  S  R  N  K   132

541  AGCGTGCCGTTCAGGGTCCAGAAGAAACAGTCACTCAAGACTGCTTGCAACTGATTGCAG   600
133   R  A  V  Q  G  P  E  E  T  V  T  Q  D  C  L  Q  L  I  A  D   152
                                                          CD-IV
```

FIG.1A

Neutrokine-α

```
601  ACAGTGAAACACCAACTATACAAAAAGGATCTTACACATTTGTTCCATGGCTTCTCAGCT  660
153    S  E  T  P  T  I  Q  K  G  S  Y  T  F  V  P  W  L  L  S  F  172
                                                  CD-V

661  TTAAAAGGGGAAGTGCCCTAGAAGAAAAAGAGAATAAAATATTGGTCAAAGAAACTGGTT  720
173    K  R  G  S  A  L  E  E  K  E  N  K  I  L  V  K  E  T  G  Y  192
          CD-V                                    CD-VI

721  ACTTTTTTATATATGGTCAGGTTTTATATACTGATAAGACCTACGCCATGGGACATCTAA  780
193    F  F  I  Y  G  Q  V  L  Y  T  D  K  T  Y  A  M  G  H  L  I  212
                CD-VI                                      CD-VII

781  TTCAGAGGAAGAAGGTCCATGTCTTTGGGGATGAATTGAGTCTGGTGACTTTGTTTCGAT  840
213    Q  R  K  K  V  H  V  F  G  D  E  L  S  L  V  T  L  F  R  C  232
             CD-VII                                 CD-VIII
                                  #
841  GTATTCAAAATATGCCTGAAACACTACCCAATAATTCCTGCTATTCAGCTGGCATTGCAA  900
233    I  Q  N  M  P  E  T  L  P  N  N  S  C  Y  S  A  G  I  A  K  252
          CD-VIII                          CD-IX

901  AACTGGAAGAAGGAGATGAACTCCAACTTGCAATACCAAGAGAAAATGCACAAATATCAC  960
253    L  E  E  G  D  E  L  Q  L  A  I  P  R  E  N  A  Q  I  S  L  272
                      CD-X

961  TGGATGGAGATGTCACATTTTTTGGTGCATTGAAACTGCTGTGACCTACTTACACCATGT  1020
273    D  G  D  V  T  F  F  G  A  L  K  L  L                     285
                   CD-XI

1021 CTGTAGCTATTTTCCTCCCTTTCTCTGTACCTCTAAGAAGAAAGAATCTAACTGAAAATA  1080

1081 CCAAAAAAAAAAAAAAAAA  1100
```

FIG.1B

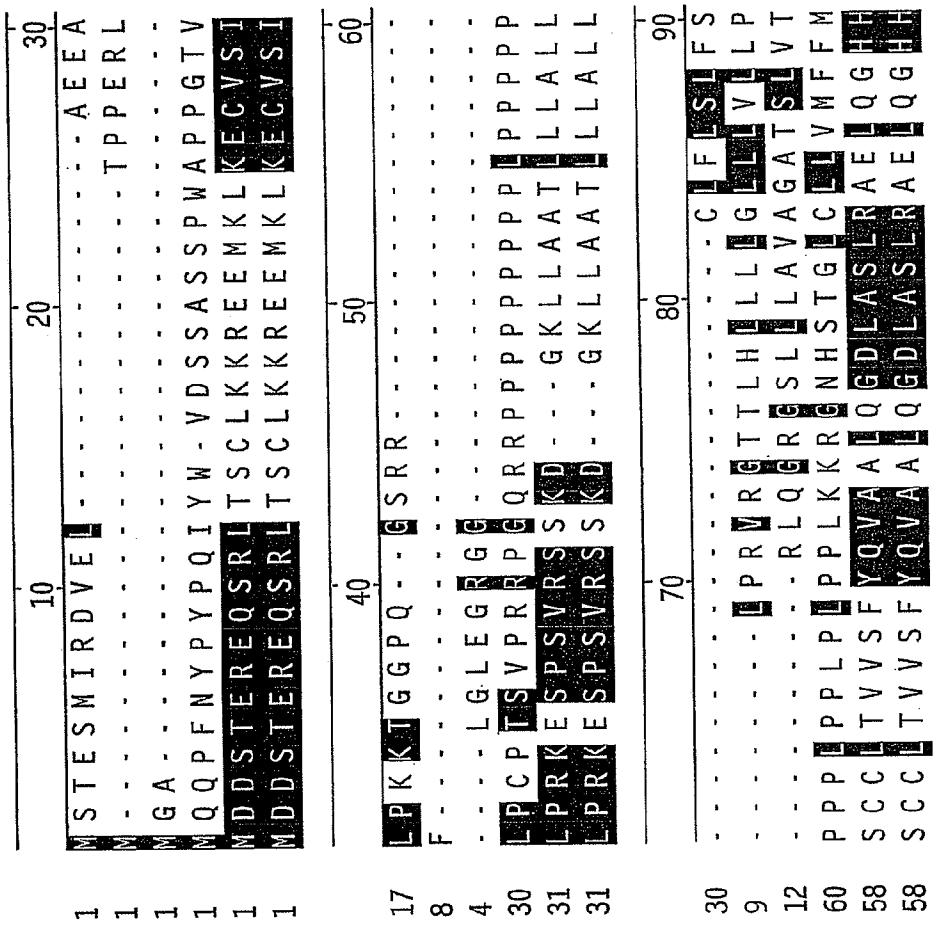
10
20
30
40
50
60
70
80
90
TNFalpha
TNFbeta
LTbeta
FasLigand
Neutrokine alpha
Neutrokine alphaSV

FIG.2A

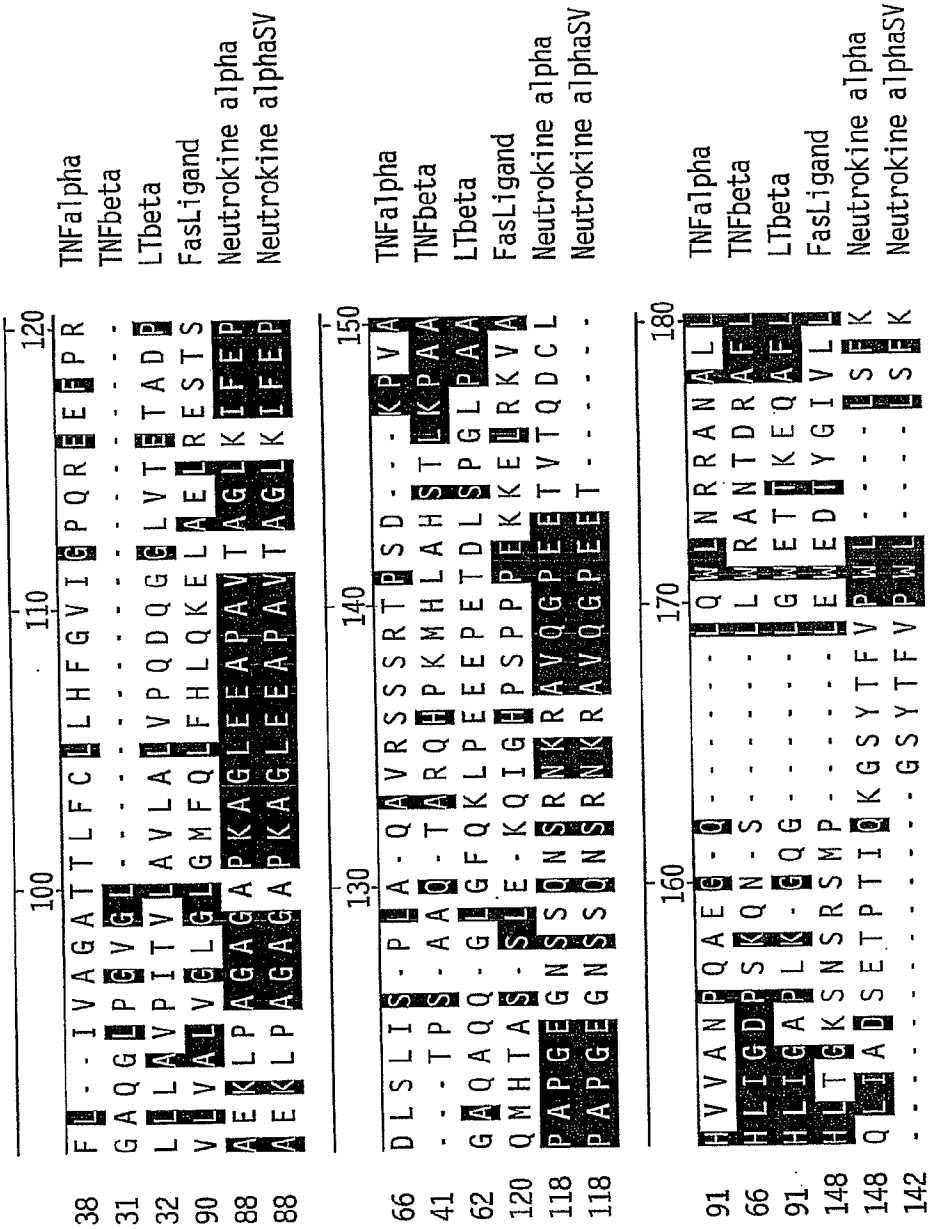
100
110
120
130
140
150
160
170
180
TNFalpha
TNFbeta
LTbeta
FasLigand
Neutrokine alpha
Neutrokine alphaSV

FIG.2B

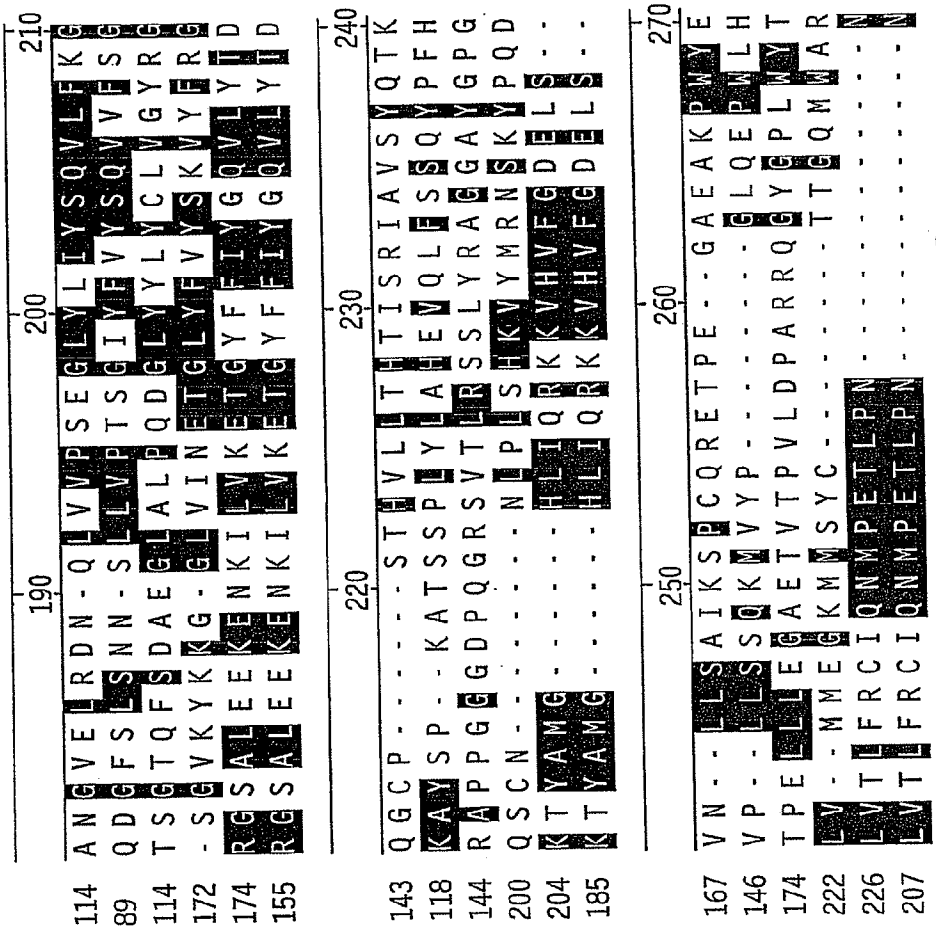
TNFalpha
TNFbeta
LTbeta
FasLigand
Neutrokine alpha
Neutrokine alphaSV
190
200
210
220
230
240
250
260
270

FIG.2C

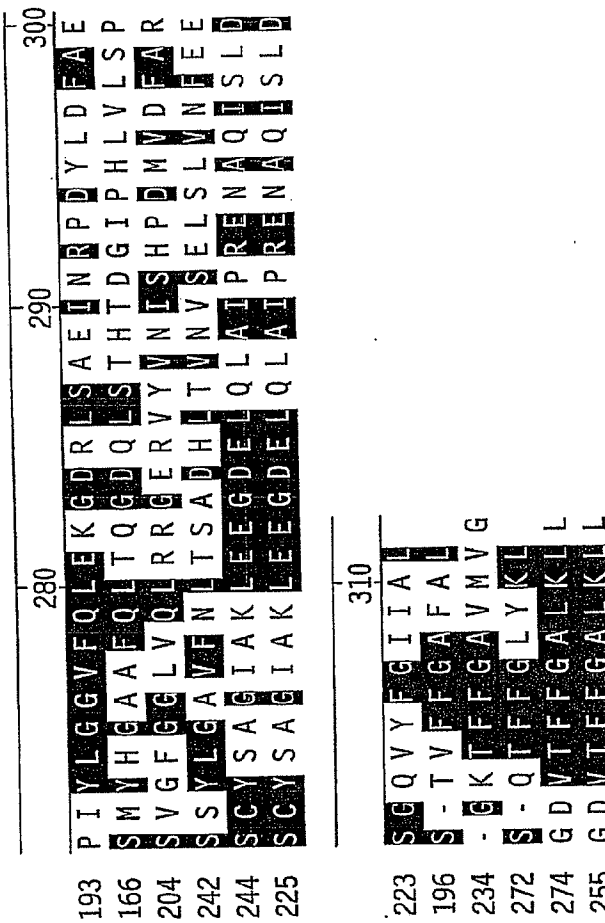
280
290
300
310
TNFalpha
TNFbeta
LTbeta
FasLigand
Neutrokine alpha
Neutrokine alphaSV
193
166
204
242
244
225
223
196
234
272
274
255

FIG.2D

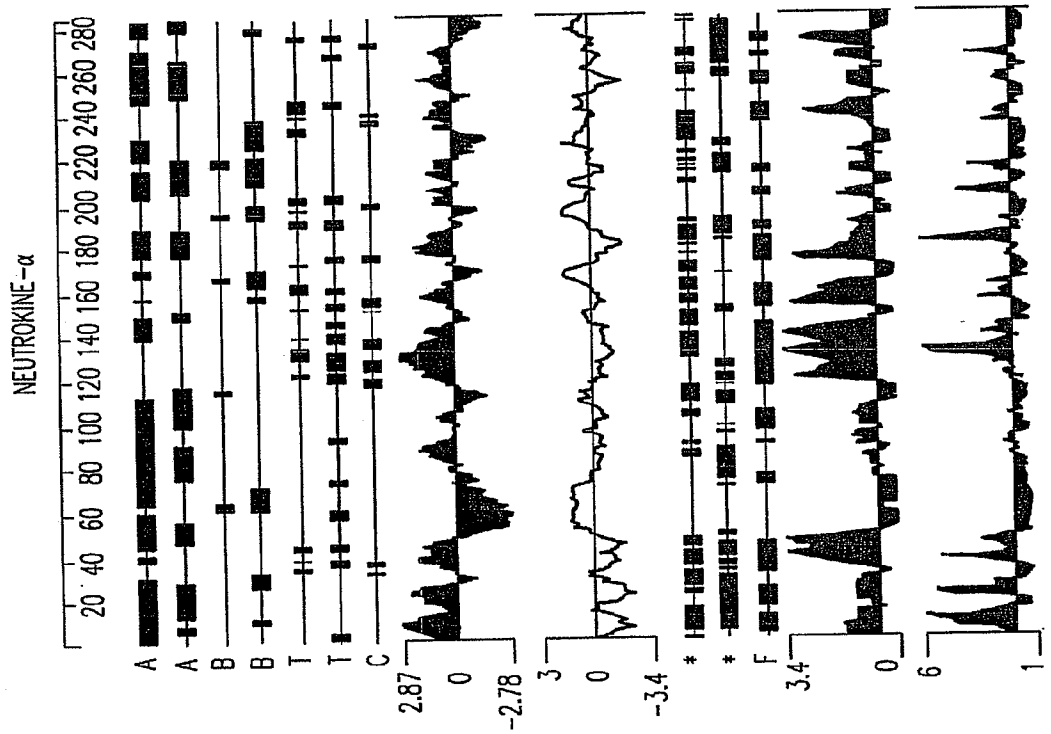
NEUTROKINE-α
20 40 60 80 100 120 140 160 180 200 220 240 260 280
A
A
B
B
T
T
C
ALPHA, REGIONS-GARNIER-ROBSON
ALPHA, REGIONS-CHOU-FASMAN
BETA, REGIONS-GARNIER-ROBSON
BETA, REGIONS-CHOU-FASMAN
TURN, REGIONS-GARNIER-ROBSON
TURN, REGIONS-CHOU-FASMAN
COIL, REGIONS-GARNIER-ROBSON
2.87
0
-2.78
HYDROPHILICITY PLOT-KYTE-DOOLITTLE
3
0
-3.4
HYDROPHOBICITY PLOT-HOPP-WOODS
*
*
F
ALPHA, AMPHIPATHIC REGIONS-EISENBERG
BETA, AMPHIPATHIC REGIONS-EISENBERG
FLEXIBLE REGIONS-KARPLUS-SCHULZ
3.4
0
ANTIGENIC INDEX-JAMESON-WOLF
6
1
SURFACE PROBABILITY PLOT-EMINI

FIG.3

```
                1                                                   50
HSOAD55R  .........A GGNTAACTCT CCTGAGGGGT GAGCCAAGCC CTGCCATGTA
HNEDU15X  ...AAATTCA GGATAACTCT CCTGAGGGGT GAGCCAAGCC CTGCCATGTA
HSLAH84R  .AATTCGGCA NAGNAAACTG GTTACTTTTT TATATATGGT CAGGTTTTAT
HLTBM08R  AATTCGGCAC GAGCAAGGCC GGCCTGGAGG AAGCTCCAGC TGTCACCGCG

               51                                                  100
HSOAD55R  GTGCACGCAG GACATCANCA A..ACACANN NNNCAGGAAA TAATCCATTC
HNEDU15X  GTGCACGCAG GACATCAACA A..ACACAGA TAACAGGAAA TGATCCATTC
HSLAH84R  ATACTGATAA GACCTACGCC ATGGGACATC TAGTTCAGAG GAAGAAGGTC
HLTBM08R  GGACTGAAAA TCTTTGAACC ACCAGCTCCA GGAGAAGGCA ACTCCAGTCA

              101                                                  150
HSOAD55R  CCTGTGGTCA CTTATTCTAA AGGCCCCAAC CTTCAAAGTT CAAGTAGTGA
HNEDU15X  CCTGTGGTCA CTTATTCTAA AGGCCCCAAC CTTCAAAGTT CAAGTAGTGA
HSLAH84R  CATGTCTTTG GGGATGAATT GAGTCTGGTG ACTTTGTTTC GATGTATTCA
HLTBM08R  GAACAGCAGA AATAAGCGTG CCGTTCAGGG TCCAGAAGAA ACAGTCACTC

              151                                                  200
HSOAD55R  TATGGATGAC TCCACAGAAA GGGAGCAGTC ACGCCTTACT TCTTGCCTTA
HNEDU15X  TATGGATGAC TCCACAGAAA GGGAGCAGTC ACGCCTTACT TCTTGCCTTA
HSLAH84R  AAATATGCCT GAAACACTAC CCAATAATTC CTGCTATTCA GCTGGCATTG
HLTBM08R  AAGACTGCTT GCAACTGNTT GCAGACAGTG AAACACCAAC TATACAAAAA

              201                                                  250
HSOAD55R  AGAAAAGAGA AGAAATGAAA CTGNAAGGAG TGTGTTTCCA TCCTCCCACG
HNEDU15X  AGAAAAGAGA AGAAATGAAA CT.GAAGGAG TGTGTTTCCA TCCTCCCACG
HSLAH84R  CAAAACTGGN AGGAAGGA.. ...GATGAAC TCCAACTTGC AATACCAGGG
HLTBM08R  GGCTCCCTTC TGNTGCCACA TTTGGGCCAA GGAATGGAGA GATTTCTTCG

              251                                                  300
HSOAD55R  GAAGGAAAGC CCCTCTNTCC GATCCTCCAA AGACGGAAAG CTGCTGGCTG
HNEDU15X  GAAGGAAAGC CCCTCTGTCC GATCCTCCAA AGACGGAAAG CTGCTGGCTG
HSLAH84R  GAAAATGCAC AATTATCACT GGGATGGAGA TGTTCACATT TTTTGGGTGC
HLTBM08R  TCTGGAAACA TTTTGCCAAA CTCTTCAGAT ACTCTTTNCT CTCTGGGAAT

              301                                                  350
HSOAD55R  CAACCTTGNT GNTGGCATTG TGTTCTTGCT GNCTCAAGGT GGTGTTNTT.
HNEDU15X  CAACCTTGCT GCTGGCACTG CTGTCTTGCT GCCTCACGGT GGTGTCTTTC
HSLAH84R  CATTGAAACT GCTGTGACCT NCTTACANCA NGTGCTGTTN GCTATTTTNC
HLTBM08R  CAAAGGAAAA TCTCTACTTA GATTNACACA TTTGTTCCCA TGGGTNTCTT

              351                                                  400
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  TACCAGGTGG CCGCCCTGCA AGGGGACCTG GCCAGCCTCC GGGCAGAGCT
HSLAH84R  CTNCCTNTTC TNTGGTAACC TCTTAGGAAG GAAGGATTCT TAACTGGGAA
HLTBM08R  AAGTTTTAAA AGGGGAGTGC CCTTAGGAGG AAAAGGGGAT AAATATTGGC
```

FIG.4A

```
                401                                                 450
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X GCAGGGCCAC  CACGCGGAGA  AGCTGCCAGC  AGGAGCAGGA  GCCCCCAAGG
HSLAH84R ATAACCCAAA  AAAANNTTAA  ANGGGTANGN  GNNANANGNG  GGGNNGTTNN
HLTBM08R CAAGGNACTG  GTTANTTTNT  AAATATGGTC  AGGTTTNTAT  ANCTGGTAGG

                451                                                 500
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X CCGGCCTGGA  GGAAGCTCCA  GCTGTCACCG  CGGGACTGAA  AATCTTTGAA
HSLAH84R CNNGNNGNNT  TTTNGGNNTA  TNTTNTNNTN  GGGNNNNGTA  AAAATGGGGC
HLTBM08R CCTCGCCATG  GGCATTNATT  CANGGNGAGG  NCNNTCTTTT  GGGNTGA...

                501                                                 550
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X CCACCAGCTC  CAGGAGAAGG  CAACTCCAGT  CAGAACAGCA  GAAATAAGCG
HSLAH84R CNANGGGGGN  TTTTT.....  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........

                551                                                 600
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X TGCCGTTCAG  GGTCCAGAAG  AAACAGTCAC  TCAAGACTGC  TTGCAACTGA
HSLAH84R ..........  ..........  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........

                601                                                 650
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X TTGCAGACAG  TGAAACACCA  ACTATACAAA  AAGGATCTTA  CACATTTGTT
HSLAH84R ..........  ..........  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........

                651                                                 700
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X CCATGGCTTC  TCAGCTTTAA  AAGGGGAAGT  GCCCTAGAAG  AAAAAGAGAA
HSLAH84R ..........  ..........  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........

                701                                                 750
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X TAAAATATTG  GTCAAAGAAA  CTGGTTACTT  TTTTATATAT  GGTCAGGTTT
HSLAH84R ..........  ..........  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........

                751                                                 800
HSOAD55R ..........  ..........  ..........  ..........  ..........
HNEDU15X TATATACTGA  TAAGACCTAC  GCCATGGGAC  ATCTAATTCA  GAGGAAGAAG
HSLAH84R ..........  ..........  ..........  ..........  ..........
HLTBM08R ..........  ..........  ..........  ..........  ..........
```

FIG.4B

```
          801                                                       850
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  GTCCATGTCT TTGGGGATGA ATTGAGTCTG GTGACTTTGT TTCGATGTAT
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          851                                                       900
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  TCAAAATATG CCTGAAACAC TACCCAATAA TTCCTGCTAT TCAGCTGGCA
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          901                                                       950
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  TTGCAAAACT GGAAGAAGGA GATGAACTCC AACTTGCAAT ACCAAGAGAA
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          951                                                       1000
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  AATGCACAAA TATCACTGGA TGGAGATGTC ACATTTTTTG GTGCATTGAA
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          1001                                                      1050
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  ACTGCTGTGA CCTACTTACA CCATGTCTGT AGCTATTTTC CTCCCTTTCT
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          1051                                                      1100
HSOAD55R  .......... .......... .......... .......... ..........
HNEDU15X  CTGTACCTCT AAGAAGAAAG AATCTAACTG AAAATACCAA AAAAAAAAAA
HSLAH84R  .......... .......... .......... .......... ..........
HLTBM08R  .......... .......... .......... .......... ..........

          1101
HSOAD55R  ......
HNEDU15X  AAAAAA
HSLAH84R  ......
HLTBM08R  ......
```

FIG.4C

Neutrokine-αSV

```
1   ATGGATGACTCCACAGAAAGGGAGCAGTCACGCCTTACTTCTTGCCTTAAGAAAAGAGAA  60
1   M  D  D  S  T  E  R  E  Q  S  R  L  T  S  C  L  K  K  R  E   20

61  GAAATGAAACTGAAGGAGTGTGTTTCCATCCTCCCACGGAAGGAAAGCCCCTCTGTCCGA  120
21  E  M  K  L  K  E  C  V  S  I  L  P  R  K  E  S  P  S  V  R   40
                                   CD-I

121 TCCTCCAAAGACGGAAAGCTGCTGGCTGCAACCTTGCTGCTGGCACTGCTGTCTTGCTGC  180
41  S  S  K  D  G  K  L  L  A  A  T  L  L  L  A  L  L  S  C  C   60
      CD-I

181 CTCACGGTGGTGTCTTTCTACCAGGTGGCCGCCCTGCAAGGGGACCTGGCCAGCCTCCGG  240
61  L  T  V  V  S  F  Y  Q  V  A  A  L  Q  G  D  L  A  S  L  R   80
                                         CD-II

241 GCAGAGCTGCAGGGCCACCACGCGGAGAAGCTGCCAGCAGGAGCAGGAGCCCCCAAGGCC  300
81  A  E  L  Q  G  H  H  A  E  K  L  P  A  G  A  G  A  P  K  A   100
    CD-II                                  CD-III

301 GGCCTGGAGGAAGCTCCAGCTGTCACCGCGGGACTGAAAATCTTTGAACCACCAGCTCCA  360
101 G  L  E  E  A  P  A  V  T  A  G  L  K  I  F  E  P  P  A  P   120
  CD-III

          #
361 GGAGAAGGCAACTCCAGTCAGAACAGCAGAAATAAGCGTGCCGTTCAGGGTCCAGAAGAA  420
121 G  E  G  N  S  S  Q  N  S  R  N  K  R  A  V  Q  G  P  E  E   140

421 ACAGGATCTTACACATTTGTTCCATGGCTTCTCAGCTTTAAAAGGGGAAGTGCCCTAGAA  480
141 T  G  S  Y  T  F  V  P  W  L  L  S  F  K  R  G  S  A  L  E   160
                                   CD-IV

481 GAAAAAGAGAATAAAATATTGGTCAAAGAAACTGGTTACTTTTTTATATATGGTCAGGTT  540
161 E  K  E  N  K  I  L  V  K  E  T  G  Y  F  F  I  Y  G  Q  V   180
  CD-IV                          CD-V

541 TTATATACTGATAAGACCTACGCCATGGGACATCTAATTCAGAGGAAGAAGGTCCATGTC  600
181 L  Y  T  D  K  T  Y  A  M  G  H  L  I  Q  R  K  K  V  H  V   200
            CD-VI                  CD-VII
```

FIG.5A

Neutrokine-αSV

```
601  TTTGGGGATGAATTGAGTCTGGTGACTTTGTTTCGATGTATTCAAAATATGCCTGAAACA  660
201  F  G  D  E  L  S  L  V  T  L  F  R  C  I  Q  N  M  P  E  T    220
     CD-VIII                    CD-VIII

661  CTACCCAATAATTCCTGCTATTCAGCTGGCATTGCAAAACTGGAAGAAGGAGATGAACTC  720
221  L  P  N  N  S  C  Y  S  A  G  I  A  K  L  E  E  G  D  E  L    240
                  CD-IX                       CD-X

721  CAACTTGCAATACCAAGAGAAAATGCACAAATATCACTGGATGGAGATGTCACATTTTTT  780
241  Q  L  A  I  P  R  E  N  A  Q  I  S  L  D  G  D  V  T  F  F    260
        CD-X                                                CD-XI

781  GGTGCATTGAAACTGCTGTGACCTACTTACACCATGTCTGTAGCTATTTTCCTCCCTTTC  840
261  G  A  L  K  L  L                                              266
        CD-XI

841  TCTGTACCTCTAAGAAGAAAGAATCTAACTGAAAATACCAAAAAAAAAAAAAAAAAAAA  900

901  AAA  903
```

FIG.5B

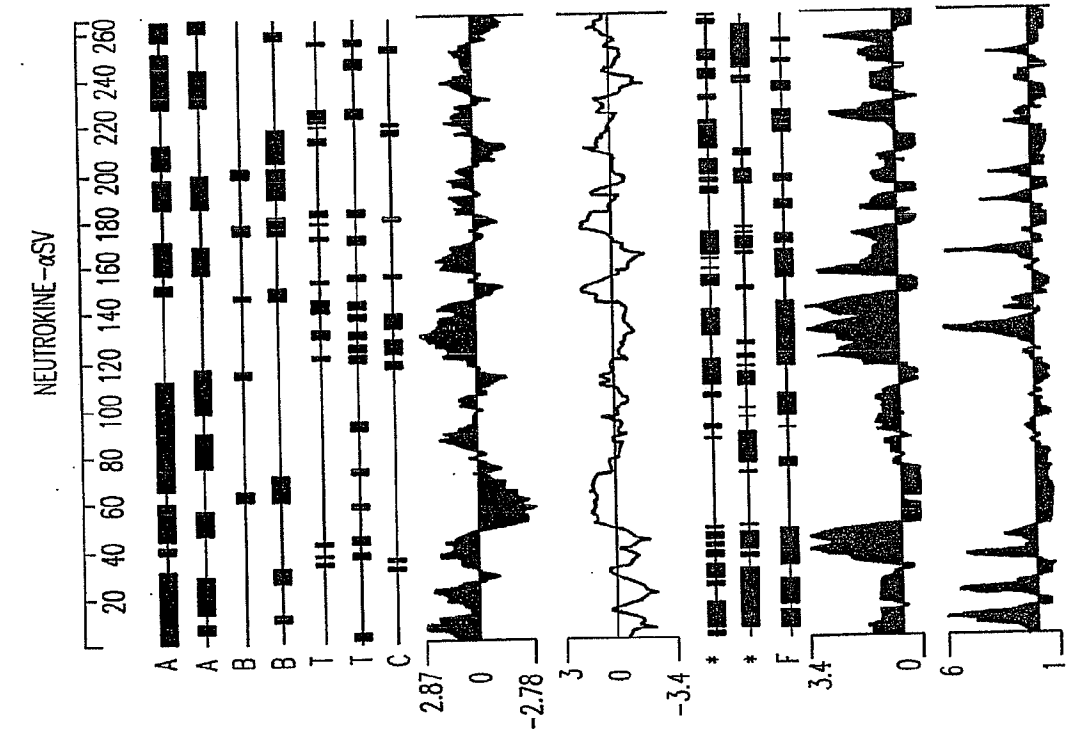
NEUTROKINE-αSV
20 40 60 80 100 120 140 160 180 200 220 240 260
A ALPHA, REGIONS-GARNIER-ROBSON
A ALPHA, REGIONS-CHOU-FASMAN
B BETA, REGIONS-GARNIER-ROBSON
B BETA, REGIONS-CHOU-FASMAN
T TURN, REGIONS-GARNIER-ROBSON
T TURN, REGIONS-CHOU-FASMAN
C COIL, REGIONS-GARNIER-ROBSON
HYDROPHILICITY PLOT-KYTE-DOOLITTLE
2.87
0
-2.78
HYDROPHOBICITY PLOT-HOPP-WOODS
3
0
-3.4
* ALPHA, AMPHIPATHIC REGIONS-EISENBERG
* BETA, AMPHIPATHIC REGIONS-EISENBERG
F FLEXIBLE REGIONS-KARPLUS-SCHULZ
3.4
0
ANTIGENIC INDEX-JAMESON-WOLF
6
1
SURFACE PROBABILITY PLOT-EMINI

FIG.6

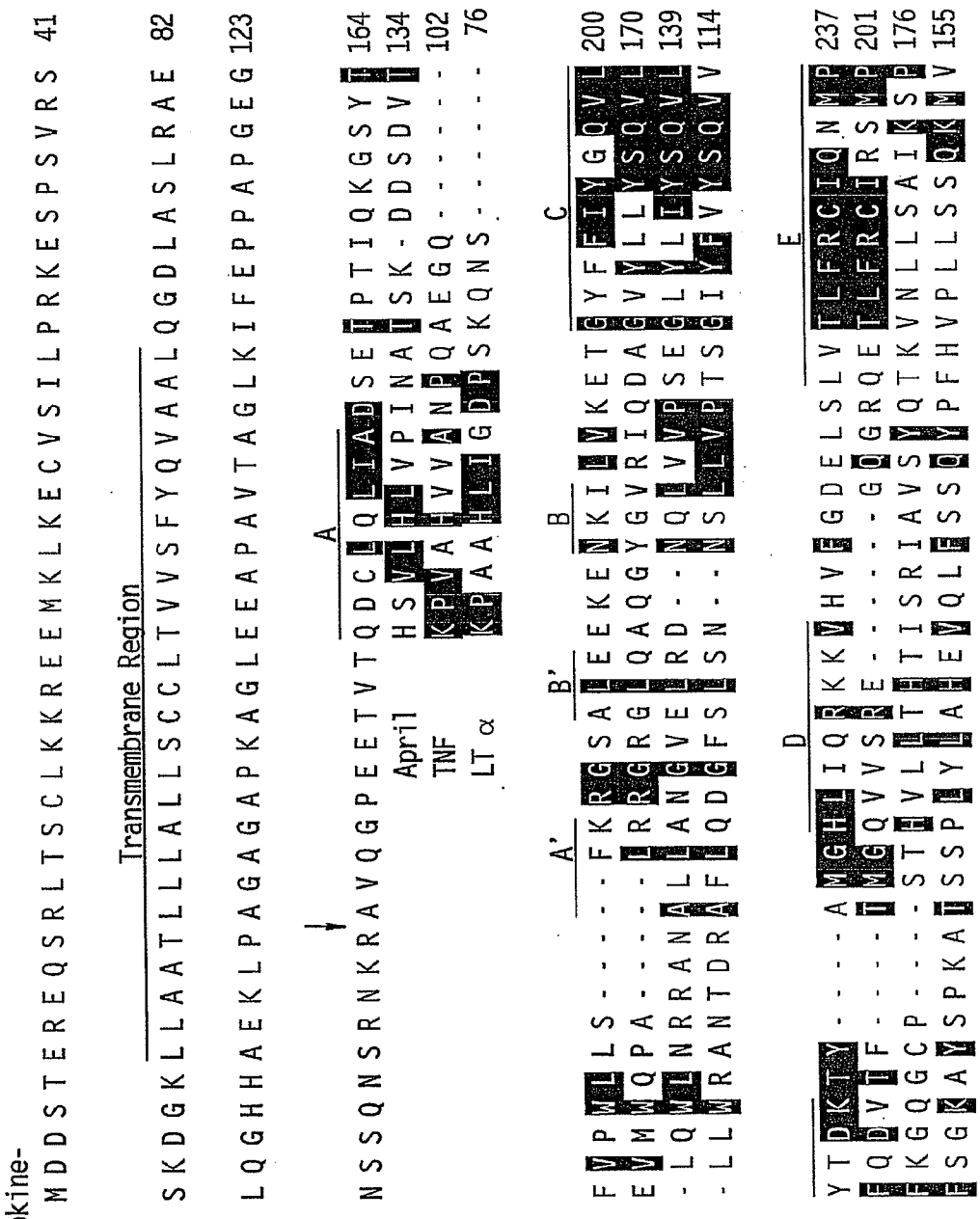
Neutrokine-
Alpha
Transmembrane Region
A
A'
B
B'
C
D
E
April
TNF
LT α

FIG.7A-1

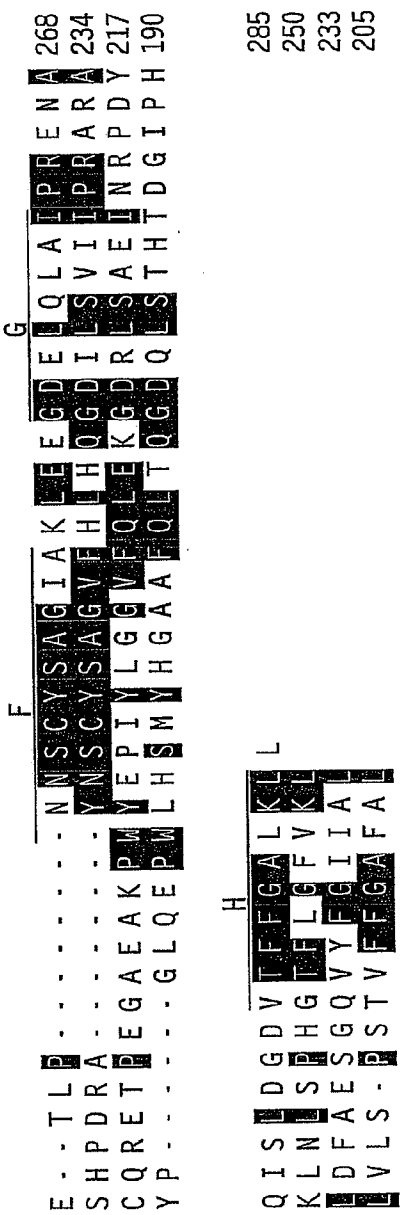

FIG.7A-2

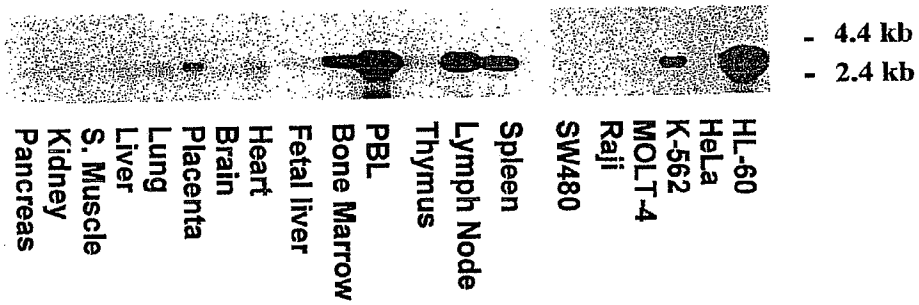
- 4.4 kb
- 2.4 kb
HL-60
HeLa
K-562
MOLT-4
Raji
SW480
Spleen
Lymph Node
Thymus
PBL
Bone Marrow
Fetal liver
Heart
Brain
Placenta
Lung
Liver
S. Muscle
Kidney
Pancreas

FIG.7B

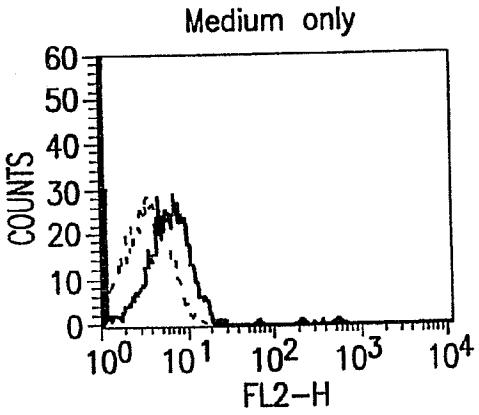

FIG.8A

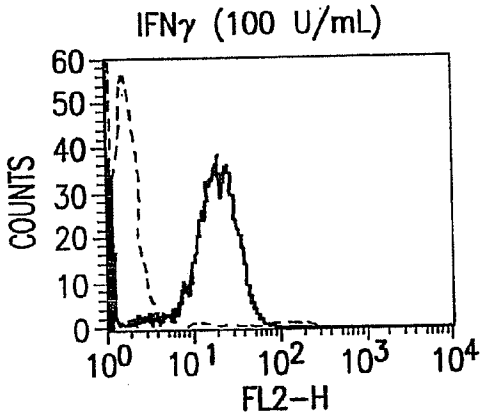

FIG.8B

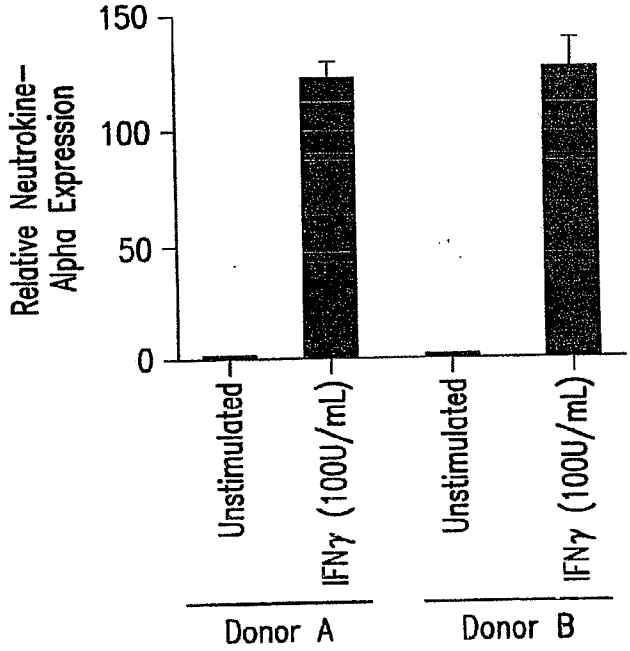

FIG.8C

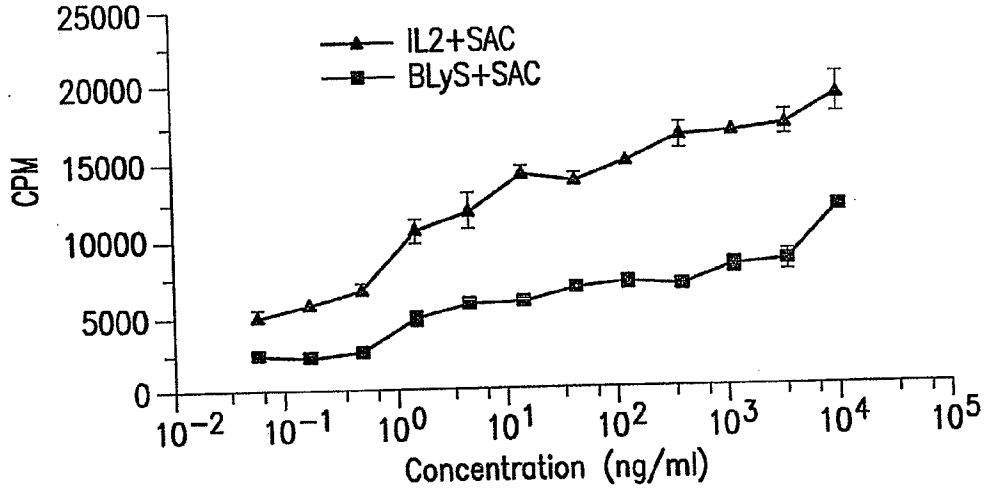
25000
20000
15000
10000
5000
0
CPM
IL2+SAC
BLyS+SAC
10^{-2}
10^{-1}
10^{0}
10^{1}
10^{2}
10^{3}
10^{4}
10^{5}
Concentration (ng/ml)

FIG. 9A

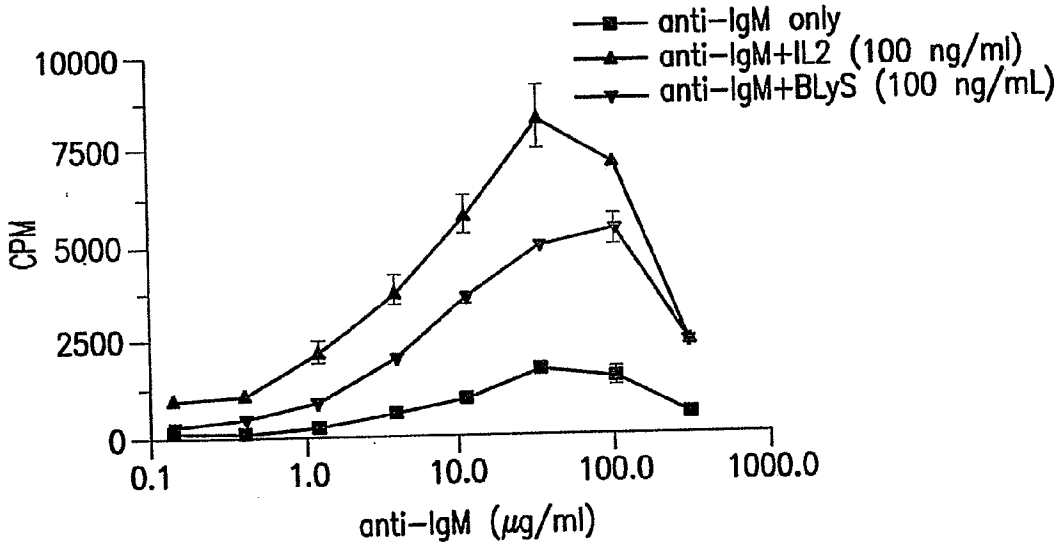
10000
7500
5000
2500
0
CPM
anti-IgM only
anti-IgM+IL2 (100 ng/ml)
anti-IgM+BLyS (100 ng/mL)
0.1
1.0
10.0
100.0
1000.0
anti-IgM (μg/ml)

FIG. 9B

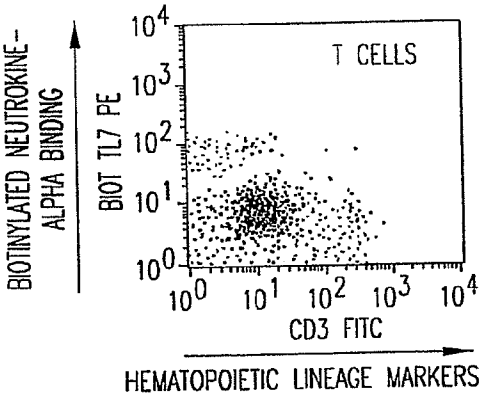

FIG.10A

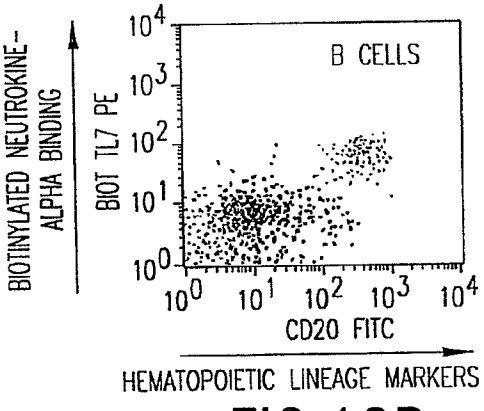

FIG.10B

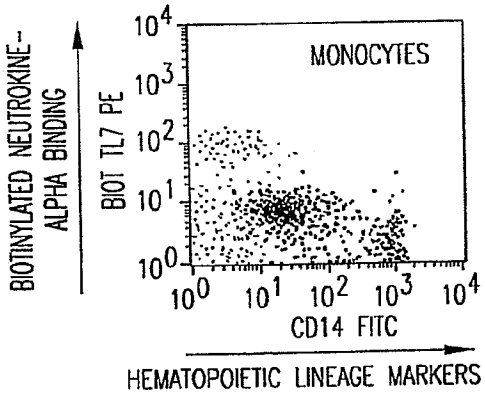

FIG.10C

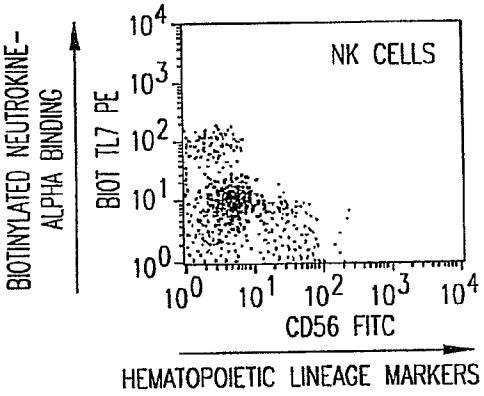

FIG.10D

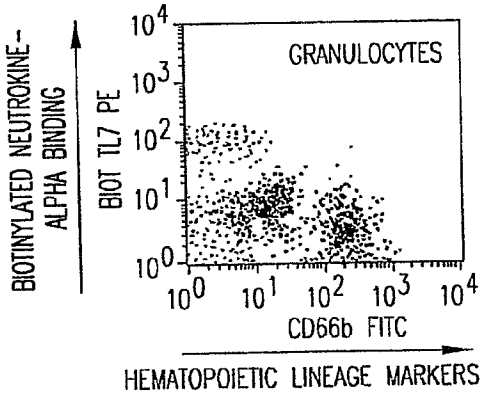

FIG.10E

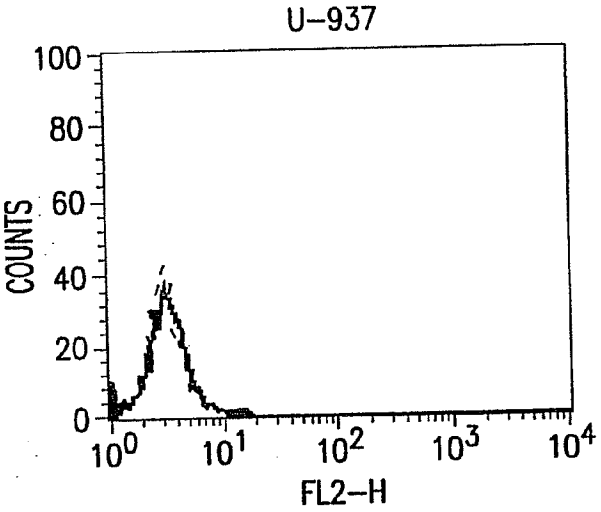

FIG.10F

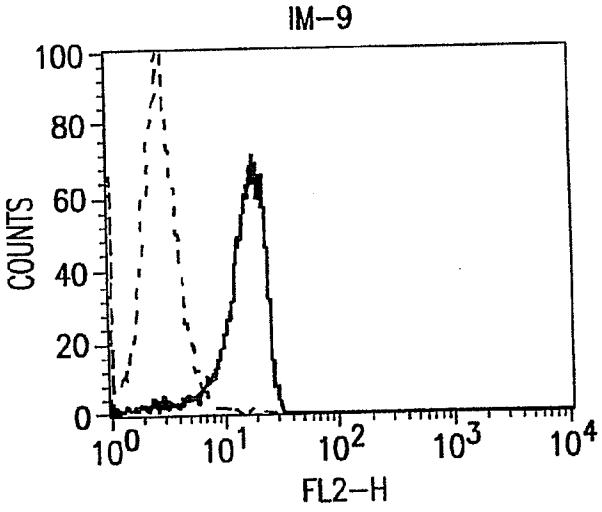

FIG.10G

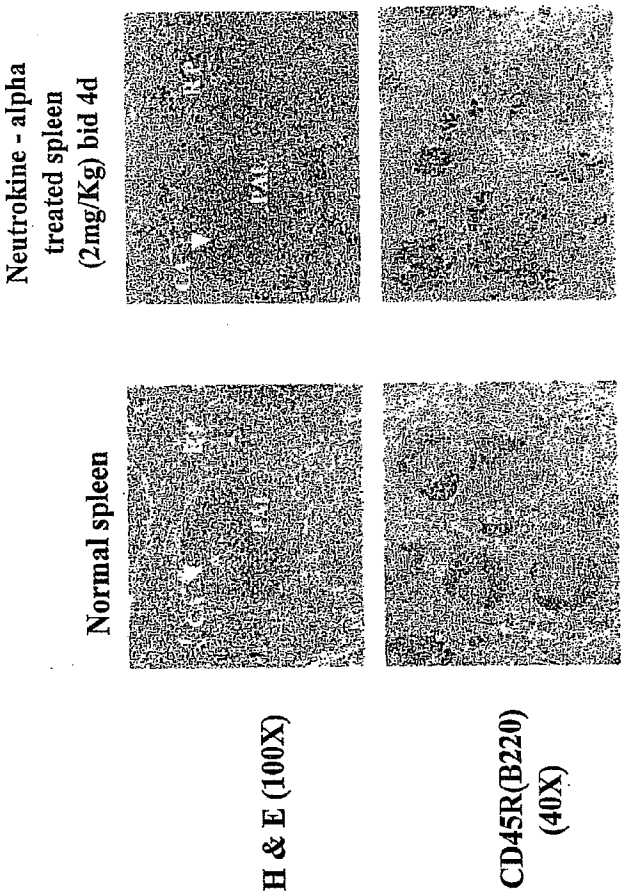
Neutrokine - alpha
treated spleen
(2mg/Kg) bid 4d
Normal spleen
H & E (100X)
CD45R(B220)
(40X)

FIG.11A

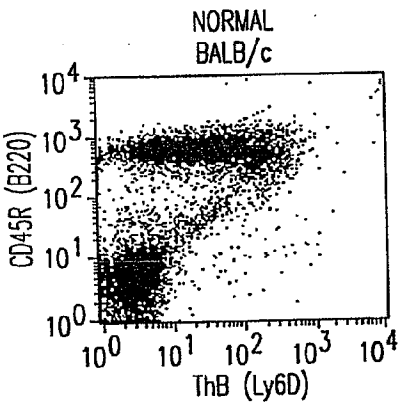

FIG. 11B

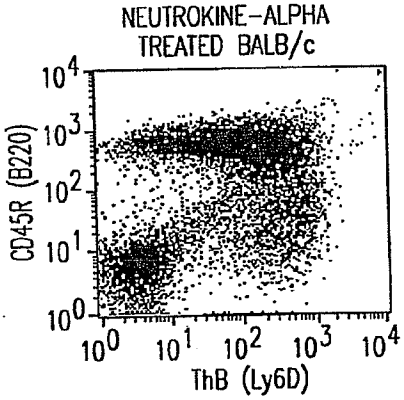

FIG. 11C

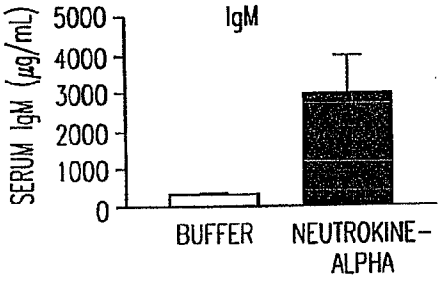

FIG. 11D

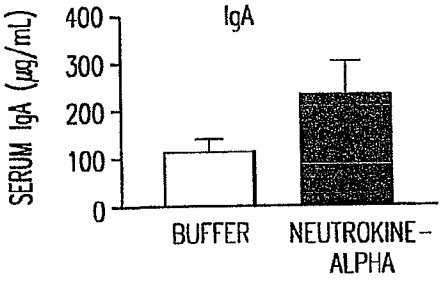

FIG. 11E

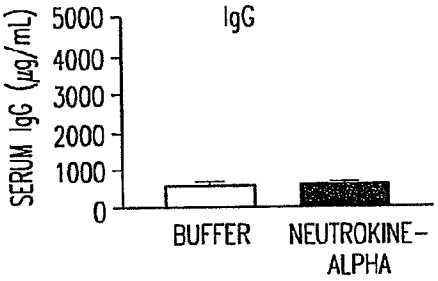

FIG. 11F